BARTLETT RESEARCH LABORATORIES

Bulletin No. 1

The European Elm Disease

A Compilation of the More Important Available Information

The Bartlett Way

PUBLISHED BY
THE F. A. BARTLETT TREE EXPERT COMPANY
STAMFORD, CONN.
1928

Aug. '28 2000

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INTRODUCTION

The experience of the last fifty years has amply demonstrated the wisdom of protecting our shade and forest trees from destructive agencies. The wide-spread blasting effect of the chestnut disease is a striking illustration of what may occur when a pernicious infection becomes established. The white pine blister rust is another unfortunate introduction.

The F. A. Bartlett Tree Expert Company has been engaged for years in practical tree protection and the Company, through its agents, has come in contact with various deadly plant diseases and destructive insect pests. When the writer was in Europe in the fall of 1927 studying shade tree conditions, he became much impressed with the serious nature of a comparatively unknown elm disease and therefore looked up available information.

This collection of papers, assembled under the direction of Mr. R. P. Marshall of the Osborn Botanical Laboratory, Yale University, is published by the Bartlett Research Laboratories as a contribution to the protection of American trees, since there is real danger of this affection becoming introduced, if it is not already established, in the country.

F. A. Bartlett.

THE TWIG WILT AND THE VASCULAR DISEASE OF THE ELM [1]

By M. B. SCHWARZ, translated by LILLIAN D. KELSEY

INTRODUCTION

For some years the elm trees in the Netherlands have suffered from a previously unknown disease. The first symptoms showed in 1919, especially on the elms in the provinces of South Holland. The disease spread rapidly in the following years, and in 1920 appeared in Rotterdam in epidemic form.

The Director of the state park, Herr D. G. Vervooren, asked help from Frl. Dr. Joh. Westerdijk, who turned further study over to me.

There was great uneasiness over this rapidly spreading disease. This is quite understandable since the elm is planted in almost all cities as a street and park tree. Also, it is frequently planted on the dikes of Holland. It is not only an ornamental tree, but the wood is technically employed.

The elm is known to be quite smoke resistant, and has here the reputation of growing readily in various soils. In Germany, however, it is considered to be one of the most particular in soil requirements according to Neger.

GENERAL APPEARANCE OF THE DISEASE

The first external appearance is the rapid wilting of the tips of the twigs. These curl and dry up. In 1920 this continued from spring into autumn and was particularly frequent in July and August. Although there was an extraordinary amount of rain, one could observe entire tops of trees and large branches dried up at the same time. The leaves died before they turned yellow. The diseased trees in the city park were immediately rigorously pruned back, and these then budded out again, healthy and fresh. By this cutting back of the diseased twigs it appeared that the wood had not yet dried, so quickly had the withering process taken place. On the contrary there was a brown ring always developed in the wood of the diseased twig in the vicinity of the cambium, with a mass of brown fluid-like excretion. The curled up tops were entirely discolored in the interior.

I, myself, cut a few trees to see how extensive the disease was. In a certain part of the city, in the vicinity of the gas-factory, there could be scarcely a tree found which did not have the brown ring in the wood, even when the tips were not entirely dried.

It may be that there are trees which are in reality diseased without showing clearly any external symptoms. It must be remembered that one who has practical knowledge of the general habit of trees, can tell whether the appearance of the ring may be expected or not by cutting.

There are certain trees with abnormally small and very pale green leaves which on cutting show all the wood browned. And there are also many cases in which one can see externally nothing abnormal on the twigs, and yet find the discolored ring in the wood.

[1] Schwarz, M. B. Das Zweigsterben der Ulmen, Trauerweiden und Pfirsichbaume, ein vergleichend—pathologische Studie. Utrecht, A. Oosthoek, pp. 7-32, 1922.

The partial brown discoloration is the primary and only sure symptom of the disease.

The browning appears not only in the wood, it appears also as a discoloration in the walls of the vascular bundles. The bark is always perfectly sound. By counting one may see in which year the disease must have entered into the tree. I shall later refer to this.

The browning was found in the rings from 1918. The disease continued much longer than one had thought in the beginning, but appeared generally externally in 1919, and in the years 1920-1921 it became epidemic. The ring is not always closed. The discolored vasculars in the ring are visible in the very youngest wood. In more advanced cases the section has a much wider unbroken discoloration in the ring of the year, although the young wood is sound.

The spread of the disease in the vascular bundles is best observed on newly infected twigs which have had the bark removed. The upper surface of the young wood shows brown longitudinal streaks. In a section of such a twig the diseased vasculars appear as brown spots. Shortly after the attack the brown vascular bundles are infrequent; later the number increases until finally not a single healthy bundle can be found. When one takes the bark from such a twig the ring of the diseased wood shows dark through the cambium. A section of a twig which was found in such a state shows a discolored closed ring.

When one follows the long streaks of the diseased twig in a new infection, it appears that further downward the discoloration generally grows fainter, and finally disappears entirely. This phenomenon is worthy of study because it signifies that the disease begins above and extends downward. I never saw it in any other way.

One may follow the browning of an older twig upward, and see how the ring is formed on the one-year twig through the anastomoses of the vascular bundles. When the discoloration is followed into the latter, one comes finally to the leaves on which the browning begins to show.

As all the branches of a twig come together in the trunk, so one can see the discoloration of the vascular bundles from the twigs finally unite until they appear as a single ring in the wood.

The ring is not always similarly developed. One may often differentiate darker and broader rings nearer weaker places. The first originates from a badly diseased part of the crown.

One may also follow the ring in its upward growth. When the twigs are very much diseased, the discoloration reaches to the furthest branching of the roots, and on the sides where the crown has been the worst infected the discoloration is furthest extended.

THE SOURCE OF THE DISEASE

As I have stated above the appearance of the twig disease of the elms in Rotterdam was especially serious in a certain part of the city. This was in the neighborhood of the gas factory. The moist ground was often heated there and ventilation was limited.

On felling the nearly dead trees the ground was found to be extremely acid and rich in *Armillaria mellea* (Vahl) Quel. The supporting parts were

entirely covered with fungi. The roots of the elms looked healthy externally, but almost all showed the browning of the wood.

According to these circumstances one might believe the source of the disease was to be looked for in the soil, although one saw cases in which the wood discoloration was limited only to the crown. On this account it has been set forth that the disease spreads downward from the twigs. The source is also to be looked for in the crown itself.

Outwardly one does not see any fungus on the diseased and lately killed twigs which could be held as the responsible agent. Locally there appeared on the dead branches numerous red mildews which without doubt did a great deal of injury, but cannot be considered as the causal agent because there are many diseased elms which are free from mildew.

There remained therefore the study as to whether an organism could be cultured from the discolored wood. So in the manner before described I placed a sterilized piece of the diseased wood on cherry agar in a petri dish.

At the beginning of the experiment several different fungi developed in the petri dish, especially Fusarium. They always established themselves on the side of the twigs which had been longest dead. One could isolate them if the browned wood from the interior of the twig was used, but not from the infected tips of the twigs. In these cases a certain fungus always developed and this only from the discolored vascular bundles. The Fusarium and other fungi did not further appear, and they were not in the interior of the wood.

I have isolated many hundreds and found the fungus only from the discolored wood, never from the normal. It is, however, immaterial whether the brown wood used was from the twigs or the roots.

Artificial infection experiments have demonstrated that this fungus is the exciting cause of the elm twig death. It is found in the wood. The result of it in the vascular bundles is such that the walls so infected are destroyed.

MYCOLOGICAL

Graphium ulmi nov. spec.

On cherry agar the elm fungus developed as a white filamentous mycelium which spread out from the wood in concentric rings day and night. By microscopic examination it appeared that one had to do with a Hymenomycete. The mycelium is rich in plasma and forked, occasionally branched. The hyphae terminate in conidiophores on which many spores are situated, held together by a slime and showing as small heads. They easily float in water.

The small spore-heads appear under the microscope as clear, glistening droplets on the snow-white mycelium. The microscopic appearance is many formed. All the forms are shown in Fig. 1. The hyphae may be slightly swollen at the point and carry numerous larger sterigma-like forms which cut off the smaller spores. Forms of different sizes are found, in one where spores, sterigmata and all intermediate forms between these are shown.

It is this great variability which makes it impossible to classify this form in a particular genus. It is somewhat like Cephalosporium, but does not fully correspond therewith. It is very unfortunate that the fungus cannot be classified in its mycelial stage; for convenience I give this form the name A (6 in

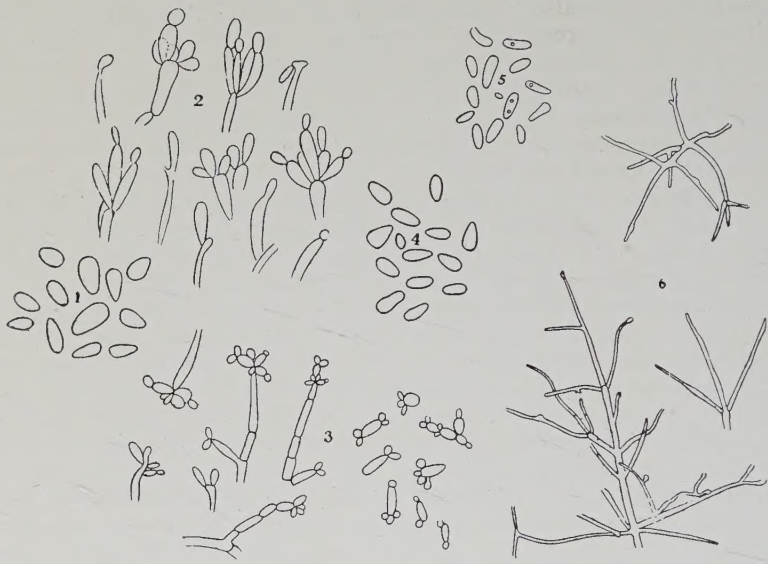

FIG. 1. THE DIFFERENT FORMS OF THE GRAPHIUMS IN CULTURE.

1.	Spores from the A-stage grown in cherry agar	1200 X
2.	Spore-carrying hyphae from the A-stage grown in cherry agar	1200 X
3.	Yeast-like spore development. B-stage from cherry juice	525 X
4.	Coremia spores	1200 X
5.	Spores from A-stage grown on rice	1200 X
6.	Hyphae from A-stage grown on rice	525 X

Fig. 1). In old cultures round to pear-shaped chlamydospores are produced in the hyphae.

Often the cultures in the dish are very sticky, and it appears as if contamination by bacteria had occurred. By microscopic examination one finds in these cases a yeast-like organism. Yeast frequently appears as a contamination, especially in the isolation of fungi from diseased wood. I had not sufficiently observed this in the early stages, but the culture experiment showed that the A-stage under certain circumstances can pass into the yeast-like form, which I will designate as B. I shall consider this question in the following chapters. Here I will only state that the B-stage (3 in Fig. 1) does not show an isolation when the pieces of wood are dried, with sterile filter paper, before they are placed in the culture medium.

After a few days there appear in the petri-dishes on the wood and later on the agar itself between the hyphae, dark brown to black stemmed coremia with large, clear spore-heads held together by a slime.

In the beginning it seemed probable that the coremia did not belong to the white-threaded mycelium, yet through culture experiments, such was shown to be the case. The variant A shows only a deeper development stage of the coremia fungus of the family Stilbaceae. I have not only germinated many hundreds of coremia spores and always seen the characteristic white A

develop, but there also resulted, on the other hand, from hundreds of isolated germinating A spores the same A colonies in which, after the lapse of a short time, the coremia were developed. Also in cover-glass cultures the same fungus always resulted from a single spore, whether from the small head or from the A-stage.

In cultures on definite nutrient materials, for example, carrots, one may see the transition form from the usual mycelium to the head. One finds the peculiar branching of the spore carrying hyphae from the white mycelium in the head as soon as the extraneous spore mass is removed by pressure. It then shows much more clearly the brown stipe which has thick-walled cylindrical celled projecting hyphae. The threads stretch out from one another, are hyaline, septate and many times branched. They carry inverted egg-shaped conidia with visible oil-drops at the apex. The differentiation from the spore carriers of the A stage is that the sterigma is not present. The spore mass is more homogenous.

The fungus belongs to the genus Graphium, and corresponds with none of the described species. In section, the spores measure $3.25 \times 1.71\mu$, the borders are $2\text{-}5 \times 1\text{-}3\mu$.

According to the description in Rabenhorst, considering the size of the spores, there is a Graphium species which is somewhat similar. This is *Graphium pencillioides* Corda, with spores of $4\text{-}5 \times 1.5\mu$. There were, however, unbranched conidiophores described. This species is not identical because the conidiophores possess important differences.

In regard to *Graphium desmazieri* Sacc., here the spores may correspond with those of the elm fungus, for they measure $3\text{-}5 \times 1.5\text{-}2.5\mu$. However, the mycelium is smoke colored and as a particular difference the conidiophores have on their tips small bent teeth.

As the last comparative species, *Graphium rigida* Corda with spores from $2.5\text{-}4 \times 1.5\text{-}2\mu$, must not be overlooked. Here again the unbranched conidiophores make it impossible to identify it with the elm Graphium.

I may further state that a fungus has come to me from the Botanical Museum in Berlin which was marked as *Graphium rigidum,* although it had branched conidiophores. The substratum on which it is located is, according to the exsicatti, decayed wood. I mention the fungus here only because it is very similar to the elm Graphium.

The fact that the Graphium which has been cultured from the diseased elm is unconnected with any described species has led me to name it *Graphium ulmi,* nov. spec.

The fungus appears in 3 forms:

1. The mycelium stage which I designate A
2. The yeast-like form which I designate as B
3. The stilbacean stage. From this form I have illustrated the fungus as *Graphium ulmi*

GRAPHIUM ULMI NOV. SPEC.

Description: Coremia (gesellig?) to 1500μ high.

Stalk unbranched, smooth, up to 1200μ long and 120μ thick, of smoke-colored parallel extending hyphae which form cylindrical thick-walled cells. The stipe becomes clearer above, branching out into several branched, hyaline, septate, conidiophores which are up

to 30μ long and 2μ thick. They develop a small head which is round, hyaline but in a dry condition, opaque and yellow up to 350μ in section.

Conidia Acrogenous, hyaline, inverted egg-shaped with many visible oil drops, and developed through slime into small heads. The normal size is 3.25 x 1.71μ, the range being 2.5 x 1.3μ. These measures refer to coremia which have grown on wood.

As a secondary form may be mentioned white filamentous mycelium with many forked branching conidiophores which are slightly swollen at the tip, and with many sterigma-like developments. The sterigma are very variable in size, and unite one-celled, hyaline, inverted egg-shaped conidia having many visible oil-drops.

Conidia and sterigma are held close together by slime, and show as small heads. On cherry agar the spores measure 2.5-7 x 1.4μ, the medium size is 4 x 2μ. On different artificial nutrient bases yeast-like developments often occur. Old cultures have chlamydospores of various forms, round to pear shape.

Parasite on elm in many places in the Netherlands.

CULTURE EXPERIMENTS

In order to further study the relationship of *Graphium ulmi,* I undertook a number of cultures on varying nutrients. The tubes were similarly inoculated, using a sterilized hook or loop with spore emulsion of the A-stage, which was grown on cherry agar. The various data are shown in the table. The differences are very great. They demonstrate twigs, and especially potato stalks, as very good nutrient media. Lupine stems are not suitable. This is more remarkable because one has elsewhere used it efficiently as a medium for twig fungus culture.

The culture on rice is differentiated from the others by its dark color. It is often seen that the most intensive color of a fungus is developed on rice. It is unknown just why this is the fact. It is not the starch content, for lima beans and oat agar show no especially strong color.

Graphium does not grow luxuriantly on a nutrient base which contains sugar only as a carbonic acid ingredient, as is shown in the table.

The difference between the malt agar and malt gelatine culture is striking. The latter is rich in the filamentous white mycelium. The former shows only a heavy yellow, yeast-like mass. It makes very little difference whether one uses the loop method or inoculates with the mycelium, at least in the beginning, as in the first case there is obtained only yeast-like spore residue; in the second place there is only a scanty, sticky mycelium development.

It appears here, as Appel and Wollenweber have described, that spore-cultures almost always develop from spores; from mycelium, only mycelial cultures develop.

In the yeast-like cultures, growth like *Graphium tubercularum* is developed almost entirely without mycelium. Microscopic examination shows the yeast-like spore residue as a mass in which strongly swollen sterigma show in the foreground. Between these are seen the thin, not largely developed spores, which are more numerous than the sterigma. The hyphae appear seldom, and are developed as short, swollen, cylindrical cells. All the sterigma, spores and hyphae are rich in fats, which show in large and small drops.

The swollen sterigma begin, when placed in a cover-glass, to send off yeast-like conidia. The different forms are shown in 3, Fig. 1. The conidia may develop on all sides.

The cultures on carrots are differentiated by the branched initial stage of the stipes. They are often not fully unfolded, and one may find undeveloped

coremia which show the fine transition from the A-stage of the Graphium.

The stipe is solid and not smooth, because conidiophores develop irregularly.

Cellulose agar by itself cannot be employed as a nutrient material. The scanty hyphae have developed at the expense of the cellulose and the impurities in the agar. On the whole I will state that potato stems and tips are the best nutrient media; one may obtain fruiting Graphium on this without difficulty.

Rice and cherry agar are also favorable, only the coremia development ceases quickly on prolonged culture. The A-stage, however, grows well for some time.

I do not recommend such nutrients as malt, oats, banana agar and beans, on account of their yeast-like growths.

In general the size of the fungus spores is largely dependent upon the influence of the different nutrient media.

In order to make this perfectly clear and so that it may be measured, I have represented in curves the length and breadth of about 200 spores from different cultures. Fig. 2 shows width and length of 200 Graphium spores from a coremium which was grown from diseased wood. The peak of the spore lengths is 3 and for the width 1.5μ. The analogous course for both curves is striking. In many fungi the width is variable and also the length, only the latter must always correspond. This is not the case in *Graphium ulmi*, and one may use both.

The curves in Fig. 3 are from coremia cultured in potato stems. The spores are larger in section, the peak of the length and breadth lie between 4μ and 2μ. The course is very irregular.

From a culture from cherry agar after isolation from diseased wood, the coremia spores were measured, and the curve (Fig. 4) is compared with the spores from the same culture, only from the very variable A-stage, Fig. 5.

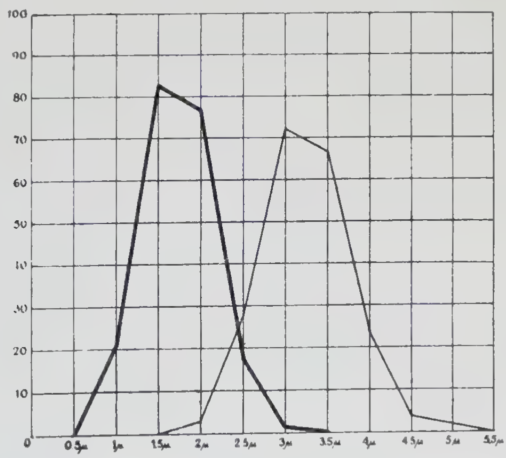

FIG. 2. 200 of coremia Graphium spores grown on wood. Heavy lines indicate width. Light lines length. The number of spores is represented vertically.

Table I. Culture Record of Graphium ulmi

Specimen	*Color*	*Mycelium Development*	*Coremia*	*Spore Appearance*
Elm Twig	White	Feathery, insignificant, only on cut surface	Here and there developed in bunches. Many on cut surface of twigs	Opaque Yellowish
Maple Twig	White	Feathery, insignificant, only on cut surface	As above	Opaque Yellowish
Potato stem	....	None	Large and numerous, over the entire upper surface	Hyaline or opaque, yellowish
Lupine stem	White	Very insignificant, only on the inoculation point	None	....
Beans	Yellowish	Almost no mycelium, culture yeast-like	None	Yellowish spore groups
Carrot (Daucus carota)	Yellowish	Culture yeast-like	Numerous undeveloped outgrowing Graphium stems; few normal Coremia	Hyaline
Apple bits	White	Rough or woolly	Numerous	Hyaline or opaque, yellowish
Meat-broth agar	White	Culture yeast-like; mycelium only on the dried walls, forming rings daily.	....	....
Cherry agar	White	Mycelium forming rings daily. Culture yeast-like in the middle.	Numerous	Hyaline or opaque, yellowish
Banana agar	Yellowish	Culture yeast-like	....	Yellowish

Table I (*continued*)

Specimen	*Color*	*Mycelium Development*	*Coremia*	*Spore Appearance*
Oatmalt agar	White	Insignificant	Few	White to opaque yellow spore groups
Lima bean agar (Phaseolus lunatus)	White	Abundant, forming rings daily	Numerous	Hyaline Coremia heads or opaque yellow spore groups
Rice	Yellow to dark brown	A little white mycelium; usually the culture is yeast-like.	Numerous	Hyaline
Beer wort salep agar	Yellowish	Culture yeast-like. Mycelium only on dry edges, sparsely developed	....	Yellowish spore groups
Beer wort agar	Deep yellow	Culture all yeast-like, on the walls developing rings	....	Deep yellow spore groups
Beer wort gelatine. No liquid	White	Feathery and bushy mycelium	Beginning of stem development	....
Salep agar	White	Mycelium sparsely developed, growing in rings	Striking development	Hyaline or opaque
Mannite agar	Hyaline	Insignificant, yeast-like	....	....
Saccharose agar 4%	Hyaline	Yeast-like, scanty growth	....	....
Cellulose agar	Hyaline	Scarcely perceptible, yeast-like growth	....	....

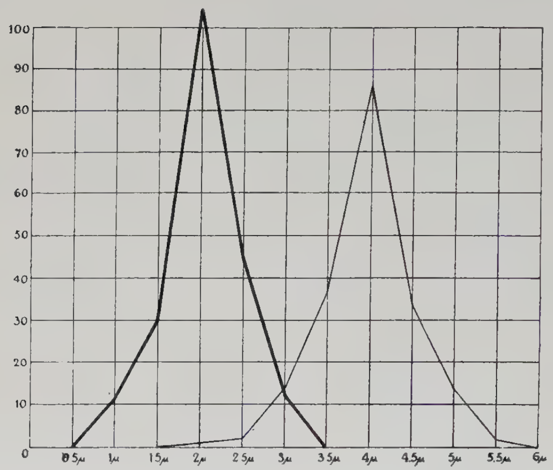

FIG. 3. 200 of coremia Graphium spores grown on potato-stalks. Heavy lines indicate width, light lines length. The number of spores is represented vertically.

It shows here that the curves of the coremia spores have great similarity to the coremia spores from the natural substratum (diseased wood). The peaks are here 3 and 1.5μ while the A spores are between 4 and 2μ. The curves themselves are, further, not so regular as in the other cases, although length and breadth in like manner are divergent.

FURTHER ANALYSIS OF THE OBSERVATIONS IN NATURE

As I have stated, the disease is not always externally visible. The wood browning is the only criterion of attack, for it is the discolored vasculars which always contain the parasites.

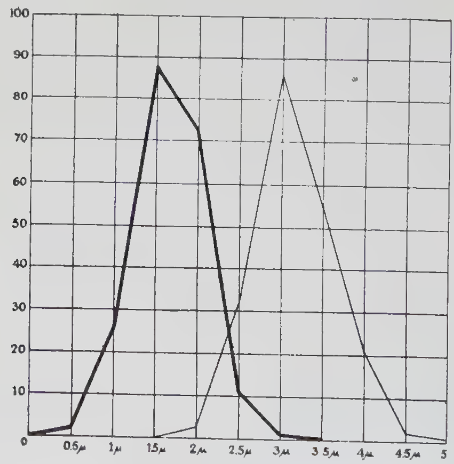

FIG. 4. 200 Coremia spores of Graphium grown on cherry agar. Heavy lines indicate width, light lines length. The number of spores is represented vertically.

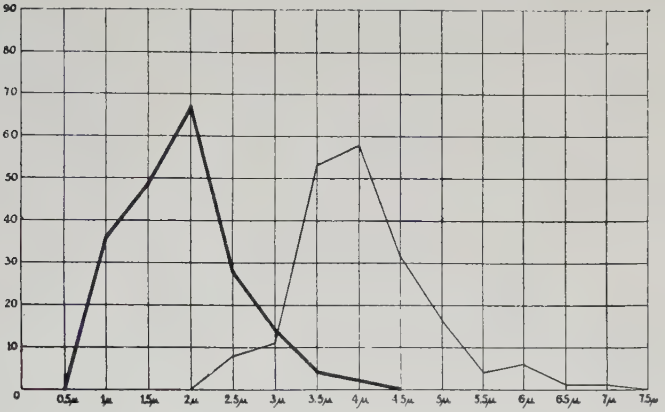

FIG. 5. 200 Coremia spores of Graphium from the A-stage grown on cherry agar. Heavy lines indicate width, light lines length. The number of spores is represented vertically.

These are not found in the ordinary wood.

Under some circumstances there occurs a spontaneous drying of the twig-tips which curl and show the characteristics of a badly diseased elm. This phenomenon was especially frequent in the summer of the year 1920 when the elm disease was so general. In the summer of 1921 another phenomenon came into prominence.

In the months of June and July the tops of the elms began to show yellow and red discolorations. They had an autumnal appearance; the leaves fell soon after. The bare twigs were mostly dead and dried, and had a broad dark ring except in the very youngest wood. Also the last year's tips had not become discolored in the interior; they had developed normally, however; they died early without showing any curling.

They were not infected, and the early death which occurred is to be differentiated from the spontaneous and rapid withering. It must be ascribed to the abnormal conditions.

I have already stated that in the year 1920 the months of July and August were distinguished by an unusually heavy rain-fall.

It was precisely at this time that the spontaneous wilting was most noticeable.

The summer of 1921 was unusually dry, and it is easily understandable that the already quite heavily charged water conductors under these conditions were no longer normal and the leaves gradually died. The death in the dry summer was secondary and the result of an infection which had taken place much earlier, in contradistinction to the fact the crumpled and internally discolored tips were primarily infected. I seldom found this in the year 1921. Apparently here the weather played a large role; therefore in a very wet summer the conditions for an acute invasion and rapid spread are very favorable.

The primary infections in the summer of 1921 did not fully mature, they produced only a moderately acute infection. I found signs of it in the form

of a few discolored vascular bundle strands in the one-year sprouts. These could be followed upward to the leaf-stem and began in the mid-rib. This fact is an important phenomenon because thereby the question is at once solved as to where the infection takes place in the open.

The leaves may also afford an entrance for the parasite.

The duration of time has been experimentally studied and proven. I placed a drop of the spore emulsion on a leaf which was kept moist in a petri-dish. Microscopic examination showed that the germ threads of the spores penetrated through the stomata of the leaf. The spore carriers with the spores came out again through the stomata. I got results only on the under side of the leaf. After 14 days there were numerous Graphium coremia developed on the leaf.

The experiment must be carried on in a very moist atmosphere, otherwise it will not be successful; the spores scarcely germinate and nó coremia appear.

Although *Graphium ulmi* finds its way through the leaf, it is not a typical leaf fungus. Infection cannot be discovered on the exterior of the leaf. No spots are developed, the leaf tissue dies gradually and the browning of the tissue commences first in the mid-rib as a slight streak. The fungus is easily cultured from such a leaf.

If the leaves are examined shortly after infection, it is found that the browning has extended only into the stalk. After the falling of the leaves the leaf scars have a perfectly normal appearance. By careful cutting one often finds cases in which the browning has extended through the petiole into the twig, and from there downward. The scar of such a leaf has a brown point at the location of the vascular bundle.

From the above it is easily recognized that mites by chewing the leaves and stems could bring about injury, as they would cause wounds through which parasites might easily gain entrance.

Further, leaves come into consideration, also bracts and leaf scars, as means of entrance. This fact has been demonstrated in the browning of the bundles which was found to have begun in the leaf scars. It is not possible to believe that the dry bracts themselves were infected.

The fresh leaf scars are, as has been proven by artificial infection experiments, easily infected. They do not develop thylloses in the torn tissues; later the wounded surface becomes corky. In autumn, after the leaves have fallen there is also the possibility of a general infection.

Without a doubt, after the autumn rains, the fallen diseased leaves carry over the tiny fungus spores which are easily spread and thus can infect the fresh scars. Therefore infection is possible almost through the entire year. It will twice reach its culminating point in spring and in fall, by the unfolding of and the falling of the leaves.

It is difficult to say over which period in the year wood infection occurs. One cannot externally observe the discoloration and measure the spread in the following year for when the twig is cut off, of course, infection will cease. Certainly in a one-year twig the spread had taken place in a single summer, but the exact time of the infection cannot be fixed.

Only by infection experiments is it possible to prove how rapidly browning progresses in a year. I am convinced that this depends in great measure on outside circumstances. Unfortunately I have no extended evidence on the

time, as I only undertook artificial infection experiments about the end of June last year. I give above the figures I have obtained.

The wood browning had no arresting influence on the length of the one-year shoots, as will be seen by the figures below. In October there were

5 absolutely healthy one-year shoots respectively
17-22-25-40-60 cm. long
5 entirely crumpled dead one-year shoots respectively
30-40-41-60-65 cm. long

THE ANATOMICAL CHANGES PRODUCED BY THE FUNGUS

Following the attack, the first changes appear in the wood in the form of bladder-like thylloses in the vascular bundles which are soon filled with them. Later these disappear, and the walls are then gummy, swollen, and colored brown.

The browning begins always in the vascular bundles and then spreads to the other elements of the wood, which in this stage appear as if macerated.

In order to study the fungus in the wood one needs sections of newly infected shoots. Between the thyllose-filled bundles are also a few which are not in this stage.

A high magnification shows a few thin hyphae which by their rich plasma and branching correspond with the A mycelium. [The author in his Fig. 6, Table II, reproduces a microphotograph in which the branching is clearly seen.] The mycelium in the vasculars is seen without artificial staining. When the preparation is placed for a time in glycerine-alcohol the threads grow further, and one can see them very clearly with a microscope by the aid of a reflecting prism.

Ultimately after a little practice it will be easy to discover the hyphae in the macerated wood, but the wall filaments of the injured vasculars cannot be readily distinguished.

When finally the hyphae are found it is not difficult to make a permanent microscopic preparation. The hyphae never develop a large mass which could mechanically clog the vessels. It is well, in this connection, to recognize the more or less puzzling influences of the vascular parasites in general.

The Graphium influence is certainly strong, since the single thin hyphae can produce such deep seated injuries; these must be caused by a change in the walls.

The vascular disease is differentiated from the *Verticillium albo-atrum* described by Reinke and Berth. In the Verticillium diseased stalks one finds the interior of the vessels filled with thick hyphae and chlamydospores.

The Graphium infected vessels are larger than the normal in section. This fact can be established by the use of a lens on a ring in a cross section in which the brown vessels are still individually visible. They are easily distinguished from the wide lumen which is still darker.

SUSCEPTIBILITY OF THE ELM SPECIES

Generally one finds *Ulmus campestris* L. planted everywhere in the Netherlands. In Rotterdam and its environs greater stress is laid on its

variants, *U. campestris* v. *monumentalis* Rehb., which is grafted on *U. campestris.*

U. monumentalis differs from the last named by its pyramidal compact growth, and its rougher bark which may be observed at the original grafting point. Besides, the leaf is more or less wavy, and begins to fall in November.

No difference in the susceptibility of any of these varieties has been found.

A small planting of weeping elms remained healthy. Owing to the small number of these it was impossible to positively state whether or not they are susceptible.

The weeping elms are a sport variety of *U. montana* Hemsl.; as, for example, *U. montana* var. *dampieri* Kirchn and *U. montana* var. *plumosa.*

ARTIFICIAL INFECTION EXPERIMENTS

In the winter of 1920-1921 I made infection experiments in the laboratory with cut twigs placed in water, as well as with sprouts in Cantonspark, Baarn, which had remained from spontaneous infection. The infection occurred always through wounds.

In the winter experiments I used a sterilized knife with which I made a cut in the twig down to the wood; I lifted a small bit of the bark and placed there mycelium with spores of a culture from rice.

Besides this I tried to infect the wood through the buds and dormant buds by sticking a needle through the buds deep into the wood. The mycelium was introduced into these wounds.

I closed the wounds with raffia, as also the check experiments, which were exactly the same except that no fungus infection was introduced.

By my artificial infection experiments it was not possible to produce an acute wilting and drying. I was able to produce marked wood discoloration which corresponded fully with the natural infection. This appearance was very clear in longitudinal sections. Also the Graphium mycelium could be detected in the discolored wood.

The wound closing proceeded in the normal manner from the cambium outward, and finally thin callous walls developed.

The wounds also closed in the check experiments and in cases where the infection did not take place. By cutting, however, one could see that the wood was discolored.

I designate the cases in which a discoloration appeared and by isolation developed Graphium as positive, the others negative.

In order to study the spread of the Graphium mycelium in wood or different ages I introduced spores and mycelium from rice cultures into sterilized wounds bored to different depths. Frequently I used branches several years old.

The experiments were conducted in several different ways: *a,* infection in the tree, *b,* in a small cut branch, *c,* in a log which was brought into an unheated room. Molds did not appear in any of these experiments.

The spread could be checked by examination of longitudinal sections.

The parasite grew as rapidly upward as downward. It showed a preference neither for the older nor the younger wood.

The experiments I have made do not coincide with those of Munchs. This investigation has set forth the fact that for the large number of fungi the spread in the wood is dependent upon the air and moisture thereof in a definite

way. An example is cited. When he allowed the blue-rot fungus of the coniferous trees, *Ceratostomella pini* Munch, to grow in a moist block of wood, he observed a spread of the fungus as shown in Fig. 6 *a*. In a dry block the fungus developed as in *b*, Fig. 6. [These two illustrations could not be reproduced.] The heart wood, which is always dryer than the sap-wood,

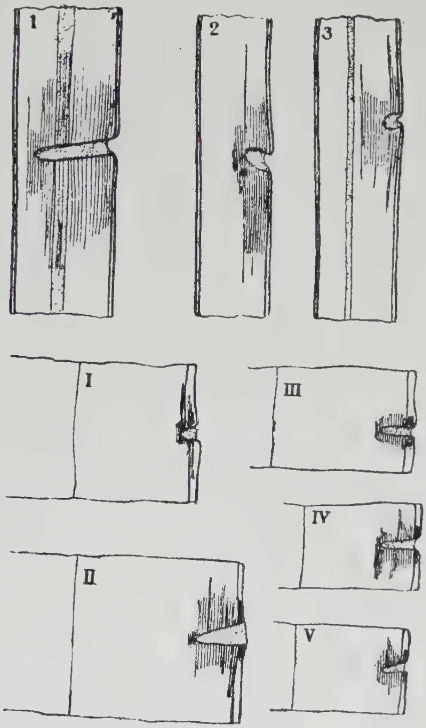

FIG. 6. 1-3, spread of fungus in moist block of wood; I-V, shows development in a stump.

has in the first example an air content which aids the development of the fungus; in the second case the air content is too small. The mycelium invades the younger wood, which although partially dried out, still holds sufficient air. The sections 1-3 of Fig. 6 are in correspondence with the experiment *a*, which was undertaken on a tree, while the sections I-V correspond to the experiment which was carried out on a stump. Table II gives the details of the infections by means of deeply bored wounds. They are positive.

The spread in small wood blocks is less extended than in the twigs on the tree. This shows a higher degree of parasitism. The result is opposed to that which Munch expected.

The bark on the side of the wound is slightly discolored. Apparently the parasite grew in through the wound-killed tissues. In experiments 1-3 the bark was too thin to show a discoloration.

Table II. Infection Experiments on Twigs of Several Years Growth

Inf. No.	Duration of Exp.	Inf. material	Diam. of twig in cm.	Depth of wound cm.	Result	Radial discoloration cm.	Discoloration in length in cm. Above	Below	Remarks
1	Jan. 12 to Sept. 15	Mycelium and spores from pure cultures from rice	1.7	1.4	Pos.	1.6	1.8 in medulla	2.8 in young wood	Exps. 1-5 were made twice on a tree. 1-3 are as 1-3 in Fig. 6. Exp. 6-8 made on a branch placed in water. Bloomed in Feb., dried in March.
2			1.1	0.35	Pos.	0.55	2	2.5″	
3			1.5	0.25	Pos.	0.65	1.4 in young wd.	1.8	
4	Jan. 12 to Oct. 20		. . .	1.2	Pos.	. . .	4	4	
5			. . .	0.2	Pos.	. . .	1.5	1.5	
6	Jan. 11 to Sept. 15		1.5	0.5	Pos.	. . .	9.5	4	In No. 6 the discoloration reached upward to cut surface. The diseased vessels lay thick under the upper surface.
7			0.6	0.2	Pos.	. . .	0.6	1	
8	Jan. 11 to July 10		0.7	0.3	Pos.	. . .	30	3	
9			4.5	0.25	Pos.	0.75	0.9 in young wood	0.6 in bark	No. 9-13 were made on a cut Block. Figs. I-V belong to Fig. 6.
10			5.5	1	Pos.	1.1	1″	1	
11			3.5	0.6	Pos.	1.1	0.3	0.2	
12			4.5	0.6	Pos.	0.65	0.7	0.6	
13			4.5	0.4	Pos.	0.45	0.6	0.4	

As far as the radial spread is concerned, the sections show that the fungus is everywhere deeper in the wood than the inoculation. This is of great importance, as thereby one can with absolute certainty know in what year the disease appears for the first time.

The artificial infections show further that the fungus grows with the same rapidity upward as downward. This appears to verify my point of view that the normal spread was from the upper part to the lower. One must not forget that these infections all took place on twigs of several years standing, while in nature only the younger twigs become infected. I can therefore only say that all observations in the open show the spread to distant points proceeded from the growing parts. So many of the dormant buds of various trees in the past summer sprouted following the drouth in August, that I checked many of the young shoots for the browning, but although the wood of the trunk was fully discolored, I found them thoroughly healthy.

The parasite grows always on the older parts in nature.

In summer I used younger parts for infection; for example twig-tips, leaf-buds, petioles and scars. I selected sprouts which were free from spontaneous infection, although these were in the same gardens with diseased trees.

I used now for inoculation a wire dipped in the spore emulsion and always introduced into wounds. These varied in the bark from small, deep cuts into the wood to wide wounds reaching only to the cambium. All were later overgrown with callus.

The leaf inoculations could not be protected by raffia, in the other cases this was used.

On the leaves the epidermis was rubbed. The infection followed when the inoculation was in the vicinity of the midrib. The wounded leaf tissues died without further result, but in the vascular bundles of the midrib there were brown streaks, which as the experiment was checked, were still visible though shrunken on the petiole. The Graphium from these places was easily cultured.

The experiments suffered great injury from the drouth; a great number had to be abandoned without results because the tree dried up. Graphium symptoms in these cases did not appear. Many leaves fell and unfortunately many of the infected.

I inoculated the leaf scars immediately after the falling of the leaves and obtained very good results.

Tables III and IV show the results on similar sprouts. It is shown clearly here that summer experiments developed better results than those made in winter.

TABLE III. INFECTION EXPERIMENTS ON SPROUTS

Inf. No.	*Duration of Exp.*	*Infection Material*	*Place of Wound*	*Result*	*Remarks*
14	Nov. 17, '20 to Aug. 12, '21	Mycelium and spores from rice culture. Isolated from roots	Bark	Pos.	
15				Pos.	Check expts. all negative
16				Neg.	
16					
17		Isolated only from stem	Dormant Buds	Pos.	Check expts. all negative
18				Neg.	
19			Buds	Neg.	
20			Bark	Neg.	
21	Dec. 22, '20 to Aug. 12, '21	The same as 17-20	Buds	Neg.	
22			Buds	Neg.	Check expts. all negative
23			Buds	Pos.	
24			Buds	Pos.	In Expt. 24 the discoloration developed 0.5 upward
25			Buds	Neg.	
26			Buds	Neg.	
27			Buds	Neg.	
28	Jan. 10, '20 to Aug. 12, '21	Like Exp. 17-20	Buds	Neg.	
29			Buds	Neg.	
30			Buds	Neg.	Check expts. all negative

Table IV. Infection Experiments with Shoots

Inf. No.	*Length of Expt.*	*Inf. Material*	*Wound point*	*Result*	*Spread of discoloration in cm.*	*Remarks*
31-32	May 28 to Aug. 5	Spore emulsion from culture in A-stage on cherry agar	2XBark	2XPos.	1	All checks negative
33			"	Pos.		Width measured only in few cases
34			"	Neg.		
35			Leaf spread	Pos.		
36			"	. . .		Leaves fallen, not checked
37-39			3XBark	3XPos.		
40			"	Neg.		
41			Leaf spread	Pos.		
42-43			2XLeaf spread	. . . .		Leaves fallen
44			"	Pos.		
45-47			3XBark	3XPos.		
48-49			2XLeaf petiole	2XPos.		
50			Bark	Doubtful		Exp. ceased on acct. drouth after 3 weeks
51			Bark	Doubtful		
52-53	July 13 to Aug. 5		2XLeaf spread	2XNeg.		
54-56			3XLeaf scar	3XPos.	0.3	In Expt. 57-76 the wounds extended to the cambium
57-60	Sept. 4 to Jan. 23		4XLeaf scar	4XPos.		
61-62			2XLeaf scar	2XPos.	0.75-0.5	
63			Bark	Pos.	2	
64	Sept. 4 to Nov. 19	Emulsion from Coremia spores	Leaf scar	Pos.		
65-67	Sept. 4 to Jan. 23		3XBark	3XPos.	0.75-1-1	
68-69			2XBark	3XNeg.		
70-71			2XLeaf scar	2XNeg.		
72-74			3XLeaf scar	3XPos.	0.5-0.2-0.75	
75-76	Sept. 4 to Nov. 10		2XBark	2XPos.		

The positive artificial infections have undoubtedly shown that the Graphium is the exciting cause of the elm wood browning. The characteristic external symptoms, through a rapid drying disease, has, however, never been produced artificially. Two reasons for this are present, *a,* the abnormal weather, and *b,* the fact that shoots or sprouts were used.

Sprouts are undoubtedly good material to work with because they neither spontaneously, or otherwise very rarely, become diseased. On the one hand there is the advantage with a check of their being no confusion between natural and artificial infection, on the other hand their slight susceptibility makes artificial infection much more difficult. I use the sprouts because they are so easy to observe, and slips or cuttings were not at my disposal.

In order to further the impression that Graphium penetrated easily into the wood, that is, into living parts of plants, I placed Graphium on freshly cut tomato and lupine stalks, without wounding the same, and infected a cut potato with the mycelium. The experiments were placed in petri dishes in a moist atmosphere. The fungus did not penetrate in any case, and the characteristic discoloration did not appear.

WEATHER INFLUENCES AND POSSIBILITIES OF CONTROL

In regard to weather I will make only a few general remarks. It obviously plays an important role in the diseases of trees. Unfortunately, I am not able to give figures in connection with the relations between weather and the elm disease, but it will be interesting to take this into account in the future.

In the present state of our biological knowledge observations which seek to analyze the correlation of living substances with the help of exact data are more and more taken into account. Certainly the studies of the special relations of growth to field and forest culture will be favored, and also the struggle between our cultivated plants and their enemies, should be first on the list. The weather has great influence in this respect and also on the animal enemies of our cultivated plants, as stated in the recently published work of Carl Borner.

The new Phaenological Society lately organized in our country can, without doubt, with its meteorological observations, be an enormous help in broadening the analyses of the weather influences and fill a great need.

While the further spread of the elm disease as affected by the influence of weather cannot now be foretold, I regard it advisable to immediately eradicate the diseased trees.

If new infections do not take place both in intensity and extension, I believe that the trees will recover very well, and with the help of the young wood may live some time. There is danger, however, of a radial spread of the fungus in the direction of the new wood, and this must be taken into account in connection with the fact that the natural path of the parasite is always away from the younger parts.

Owing to its nature, the control of the disease is not possible. To avoid the infection spray with some sort of fungicide immediately after the trees bud and before infection appears. In certain cases, for example, in the case of ornamental park trees, a good result may be attained by this means. The right time for the spraying must be ascertained.

The spread of the browning into the trunk may be prevented by an early

removal of the newly infected parts. One needs to lay no especial stress on the natural infection of the old wood by cutting. The cuts will do no injury.

LITERATURE

In 1921 there appeared from Fraulein D. Spierenburg a small publication on the elm disease. This had, however, the character of a preliminary communication rather than a publication.

Although not in direct connection with the elm disease, two works of Munch need a few words. It is identical with my investigations, notably that the genus Graphium comprises two species of wood parasites—if on fallen wood. They are the fruiting forms of two bluestain fungi.

It is well known that the bluestain of conifers is a frequent and important phenomenon treated in the work of Munch in 1907 and 1908. The only thing that interests us in these two papers is that two of the fungi considered as the cause produce as a secondary fruit form a Graphium which has not been definitely identified. From this there developed mycelium which in variability showed great resemblance to the mycelial A-stage of *Graphium ulmi*. They are the Ascomycetes, *Ceratostomella piceae* Munch, and *C. cana* Munch. Although he described and illustrated the Graphium belonging to *C. piceae* with branched conidiophores, he held it as a possibility that this coremia fungus might be identical with Penicilloides. This is again a proof, as the systematic treatment of the genus Graphium is very imperfect.

The bluestain fungi do not penetrate into the interior of fresh living wood; they do not attack the wood substance especially but live only on the contents of the parenchyma cells. They also are very different in their operation and less active. The appearance of an ascus fruiting form indicates that possibly the fungus of the elm disease may develop a further fruiting form, an ascus form.

Tubeuf ascribes the elm death in Germany in the years 1918-19-20 to the excessive blooming. He considers the phenomenon as a purely physiological one, also as a result of the need of nutrient materials.

Although I have no certainty, because he does not describe the disease in detail, I hold it not impossible that this is the same Graphium disease.

SUMMARY OF THE DISEASE APPEARANCE

The *Graphium ulmi* shows itself as a browning of the vascular bundles which is found locally in the wood and is the result of the destruction of the vascular walls.

The infection takes place by means of the leaves, stomata, wounds, and the leaf scars.

The result of an infection can, beside the browning which is always a result, be evidenced in two ways.

a, A rapid wilting of the tips of the shoots takes place which immediately curl. This appearance constitutes the acute disease phase.

b, The effect is not visible externally. The diseased twigs die only after a long time.

Both phenomena are greatly influenced by external conditions.

The disease always spreads from the point of infection to older parts. In artificial infections the parasite shows no preference, and under these circumstances spreads in all directions.

ELM BLIGHT AND ITS CAUSE, GRAPHIUM ULMI SCHWARZ[1]

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with 2 illustrations
Translated by LILLIAN D. KELSEY

The discovery of the Elm Blight (Dutch Elm Disease) 8 years ago in Holland, and afterward in Belgium, northern France and western Germany, and in 1926 in Norway, has brought out numerous monographs without, however, clearing up the question as to the real cause of the disease. In order to illustrate this, to name only a few examples, may be mentioned M. B. Schwarz in Holland, Countess von Linden and Lydia Zenneck in Germany, who held that the thread fungus, *Graphium ulmi* Schw., was responsible. Brussoff stated it was due to the fission fungus, *Micrococcus ulmi* Bruss; while Pape and Lustner regard climatic influences as responsible for the disease. Dina Spierenburg inclines to the latter opinion, and he indeed isolated fungi (*Cephalosporium acremonium, Graphium penicilloides,* etc.) from diseased elms. He however was not able to artificially produce the symptoms of Elm Blight. Nor have the first two been able to produce with fungi or bacteria convincing results, such as the wilted or dried condition on living trees. Proofs of the third opinion expressed also fail. The uncertainty of these demonstrations made to discover the primary cause of the elm disease, naturally increased the difficulty of obtaining information on the spread of the trouble, which was also confused with the Red Canker disease of trees caused by *Nectria cinnabarina* (Tode) Fr. Many of the demonstrations showed manifest errors. In order to clear up the matter it was deemed better to prove by personal observations and experiments the truth of the different opinions and statements.

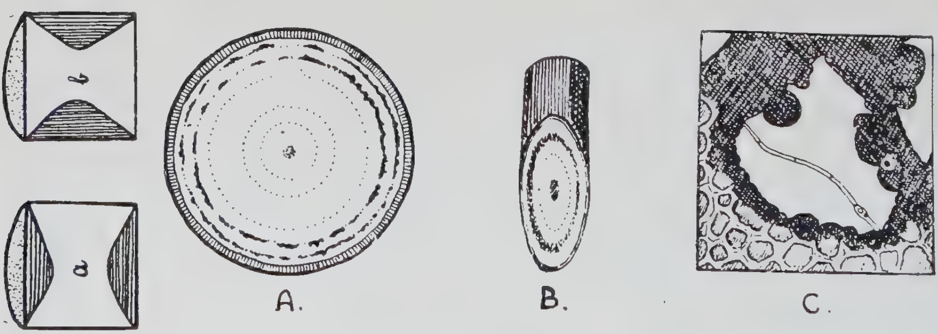

FIG. 7. Internal characters of the elm disease; *A,* represents a section of a diseased elm branch with blackish discoloration in the two outer annual rings; *B,* a two-year elm twig with very wide discolored rings; *C,* a cross section through a very much discolored vascular bundle in the wood body of a diseased elm branch with a diagonal through the tylose narrowed lumen, showing the fungus hyphae of the *G. ulmi.*

[1]Reprint from *News for German Plant Protection Service,* No. 10, 1927. Wollenweber, H. W. Das Ulmensterben und sein Erreger, *Graphium ulmi,* Schwarz. Sonderabdruck aus dem Nachrichtenblatt fur den deutschen Pflanzenschutzdienst, No . 10, 1927.

It has been incontestably proven by these experiments that the fungus, *Graphium ulmi* Schw., five weeks after inoculation of pure cultures into the wounds of one to three year old elm seedlings, produced the typical wilting phenomena and browning of the wood on all the inoculated plants, and therefore it is the true cause of the elm death. A further report will follow, in collaboration with Government Inspector Dr. Stapp, who has worked on the bacterial side of the question.

The external demonstrations of the disease are the wilting and yellowing of the foliage, defoliation and appearance of death in the branches. The occasional appearance of *Nectria cinnabarina* in the branches of trees infected with elm blight proves only that the real cause of the death of the elm often also attacks trees infected with the Nectria disease. Nectria alone produces bark decay and withering of the tips, and is easily recognized by the red pustules on the bark, and by the failure of the dark colored zones in the wood. The symptoms of death in the elm disease show from the tips of branches downward, while the browning of the wood often proceeds from below, upward. I found similar conditions in America during my studies of wilt diseases, whether caused by the Fusarium fungi in Cotton, or Verticillium fungi in Althea, etc. Young elm seedlings succumbed in a few weeks to the disease, and large trees perhaps after years, yet all the acute or chronic cases were ultimately fatal. The last year's diseased trees could be rcognized in the winter by the withered ends of the small twigs, in spring by the premature suckering on the trunk and branches, as well as the thinness of the entire foliage. Premature falling of the foliage is frequently a sign of the primary infection, but an inspection of the internal distinctive marks can only prove whether the disease can be traced back to the elm blight.

The internal indications of the disease are a brown to blackish discoloration of the vascular bundles in the trunk and branches of the infected elm, Twigs appearing quite healthy externally with heavy green foliage may show a discoloration in the wood, which when observed in a cross section of the branches may show short, indistinct flecks without a set situation, which grow into closed concentric rings of differing dimensions. Discoloration also occurs throughout the one-year wood of heavily infected twigs. Older trunks, for example, up to a mountain elm, 20 cm. thick often have a single outwardly observable dark ring in the cambium wood layers. The discoloration extends upward to the crown, but downward only to the point of cultivation. The roots were healthy in this case, a fact which may depend on the unsusceptibility of the lower parts, as in the infected elm seedlings the disease may extend into the roots. If one loosens the layers of bark from the trunk or branches in many elms, the upper surface of the wood itself seems interspersed with single brown threads which later come together into a thick net-work. In other cases the surface is perfectly normal, the browning beginning first from 1 to 2 mm. deeper in. In the bark layers the browning appears first when the branches and young shoots die.

The wood discoloration may pass in one year from the infection point in one annual ring to other rings, as these are bound together by the medulla rays. One cannot tell the year of the primary infection by the appearance of further discolored rings.

Microscopic examination easily shows the principal seat of the browning to

be the large pit vasculars in cross and longitudinal sections. Frequently the vascular bundles are partly clogged by resinous balls and bubble-like outgrowths of the walls.

The scanty, almost colorless fungus threads with transverse walls, which are scattered here and there on the walls longitudinally or in cross section, are not easily found, they are best seen, however, in longitudinal section. Occasional tangled fungus hyphae and small conidia are scattered through the lumen of the vasculars. Under oil immersion, microtome sections show this conclusively. The microscope also brings out the question of a thread fungus as a source of infection; this is in the browned wood radiations, which by placing

FIG. 8. The development of the causal organism of the elm diseases *G. ulmi; A,* Coremia branch which remained some time on an acid nutritive base, but continued nearly sterile; *B,* typical Graphium heads of various sizes which stand on black pencil-like stalks, and have bushy members with ivory or honey colored conidia droplets,—the knotted threads at the base are dark, rounded sclerotia; *C,* Conidiophores with paired or irregular branches; *D,* Cephalosporium-like conidiophores with hyphae on a slimy head, with yeast-like spores for reproduction and spread; *E,* A group of conidia connected at the ends; *F,* Conidia of the typical form (B). Figures 7 and 8 were drawn by the author and Dr. Richtus, directly from pure cultures of the disease.

the same on a nutritive base in reagent glasses is easily seen to be in a fruiting condition. The fungus is *Graphium ulmi* Schwarz. The Dutch investigator in her monograph (Utrecht 1922, pages 10-14) precisely stated this as the source of the twig dying and the vascular disease of the elm. Further particulars concerning this conclusion, together with our own findings, will be stated in another place. It is here only mentioned that the fungus belongs to the dark colored Stilbaceae, a family of the Fungi Imperfecti. Its developed forms are rounded head coremia with yellowish conidia drops on tufted, brush-like carriers, and blackish club-like stalks from long parallel fungus threads; egg to pear-shaped conidia are typical on these small heads as well as on small prostrate or half upright hyphae with regular or repeatedly whorled branching of the conidiophores, and then an outspread slimy mass in which the conidia reproduce through yeast-like sprouts; finally small rounded brown sclerotia from thick hyphae spots (size 0.045) and long outstretched club-tufted coremia (to 3 mm. high) which mostly remain sterile in contradistinction to the typical but smaller fruiting bodies which are seldom higher than 0.6 mm. In water, the conidia measure only 0.003 x 0.0016 mm. in cross section but in slime are twice as large. Mycelium tufts on the contrary are of medium size. Fruiting tubes are not observed, yet they may be found in Ceratostomella of the Ascomycete group, and are known in the group with Graphium conidia forms, among others one by Munch in studying the blue-rot of needle-woods, especially *G. persae*. Munch regards *Graphium penicilloides* Corda as a possible similar fruiting form, a form with cylindrical conidia, which is not to be confused with *Graphium ulmi,* the pear-shaped conidia of which diverge strongly therefrom.

G. ulmi grows from hundreds of over-carrying discolored wood parts, and is always exactly the same fungus from all the different localities as Duisburg, Nuremberg, Berlin, Breslau, Aachen, Dresden, etc., principally on *Ulmus montana,* but also on *U. campestris* and *U. americana.*

FACTS SHOWING GRAPHIUM ULMI AS THE CAUSE OF THE DISEASE

In July of this year 1 to 3 year old elms, maples, lindens, white thorn and poplar which had been selected from the experimental plantations at Dahlem, were inoculated with pure cultures of *Graphium ulmi* taken from different localities (Duisburg, Nuremberg). For the experiment 200 plants were used. In the inoculation experiments six other fungi which are occasionally found on elms were employed, for instance, Fusarium, Phoma, Cylindrocarpon, etc. Altogether 175 infections were made, distributed among the various plants, and fungi. The inoculations were made at the bases of the plants near the surface of the ground, in wounds, as is usual in grafting roses and orchard trees, but so that some wood was scraped under the bark in order to leave the woody vessels exposed. In these incisions the fungus in pure culture was introduced. The wounds were enveloped in wadding and tightly bound. In the first month after the inoculations almost no change appeared. With the advent of the heat wave from the end of August to the beginning of September, a few of the elms, *U. montana,* inoculated with *Graphium ulmi,* commenced to show the typical wilting symptoms, and soon all the plants in this inoculation row were diseased. The leaves became yellowish, then dried, and partly fell

off, beginning at the tip of the boughs. Internally the typical disease marks of the Elm blight showed, that is, heavy browning of the wood. The under axis had much more discolored rings, while above, often the entire wood was colored throughout. All variations from faint indistinct specks appeared. The seedlings were mostly over 1 m. high, so that the influence of the fungus in five weeks had already extended to this distance from the inoculation point. The effect of the Graphium proved marked in all cases, even to the farthest points from the inoculation, as well as elsewhere. The failure of the typical withering of the elm blight to appear, described by M. B. Schwarz in his inoculation experiments with cut twigs and branches, has been proven in full measure in our experiments, by the disease symptoms of our seedlings. (Countess von Linden and Lydia Zenneck) (Centralbl. f. Bakt. Parasit. u. Infectionskr. 11 Abt. Bd. 60, 1927, S. 351.) came to the same conclusions in their experiments.

Our ordinary experimental fungi up to the present produced no disease symptoms on elms. Also the other varieties of plants experimented with did not become infected through inoculation with Graphium nor with Fusarium, Phoma, Cylindrocarpon, etc., but it naturally cannot be said that these fungi are harmless and would so behave under other conditions.

A spontaneous outbreak of a wilting disease at our Forest School was of interest. It was frequently observed on an *Acer negundo* seedling of our experimental planting, which showed a fungus in the discolored wood vasculars like a *Verticillium dahleae* Kleb.; and indicated that we had here to do with a similar disease although of different origin. Through the negative results with *Graphium ulmi* on maple, we may therefore come to the conclusion that the maple wilt disease is truly a Verticillium disease, and the elm blight a Graphium disease. We do not know that Graphium can carry over from the elm to the beech, linden, poplar, maple and other plants, or that these plants suffer from other Graphium forms.

It is of interest that Georgevitch (Comptes rendus 1926, p. 759-761) has written of a dark grey wood discoloration of the common oak by *Ceratostomella querci,* and indeed with Graphium in conidial form whose coremia are much smaller than those in *G. ulmi.* On the other hand, it is well known that all of these ornamental forest trees may suffer from *Nectria cinnabarina* (Tode) Fr., which ascomycete produces the red pustules and twig drying, and a generally slow developing bark rot without discoloration of the wood in the annual rings, and without epidemic outbreaks under normal growing conditions and careful management.

Regarding practical control measures, we can only make conjectural remarks. It is not impossible that we may find elm varieties which are immune against Graphium. The fact that no outbreak has been noted on *Ulmus vegeta* Lindl. and also that the red elm (*U. fulva*) remains sound amongst infected *U. americana,* as well as the fact that the root system of the cultivated weeping mountain elm remained sound when the crown was already in a dying condition, at least shows that there are notable differences in susceptibility. These possibilities must be tried out by inoculation experiments on various kinds of elms. Also crossing experiments for the purpose of finding control for, and immunity to, Graphium, and experiments on the susceptibility of seedlings and seeds from different localities should be made. This, however, cannot save the infected stands, and one must question whether this object can be attained.

In cases of weak infection, observed in the early stages, the disease may perhaps be held in check, but against heavy infection into the healthy wood, there is little possibility.

Spraying with fungicidal materials, as advocated by the Dutch scientist, M. B. Schwarz, would certainly be worthless against the work of a fungus in the interior of a plant.

As to the possibility of a full protection by saturating the wood of a healthy tree, or impregnating fungus-infected wood with fungicides without injurious effect on the healthy bark, bast and cambium of living trees, we should have little faith in such a process.

As the roots of cultivated elms often remain healthy after the crown has died, sprinkling the ground with fungicidal liquids would be useless in such cases.

Other parasitic vascular wilt diseases, for example those of cotton, watermelon and beans in America, have only been controlled by selection and cultivation of immune varieties.

Measures found possible in the control of Fusarium should be sought in the case of Verticillium and Graphium.

THE DUTCH ELM DISEASE[1]

By MALCOLM WILSON

During the past few years a serious disease of the Elm has appeared in several countries in Northwest Europe, and in order to prevent the spread of this disease into England, the Ministry of Agriculture has recently issued an order prohibiting the importation of Elms from Europe into this country.

The disease was first observed in Holland in September, 1919, at Tilburg in North Brabant, and has steadily spread, until at present it is found all over that country. It was recorded over the whole of Belgium in 1921, and in the same year in France, north of the Seine. In 1924, it was found in Germany, in Rhineland, Westphalia, Bremen, Nurenberg, Potsdam and Bonn, and at Aix la Chapelle in 1925. Diseased Elms, showing exactly the same symptoms as those of the Dutch Elm disease, were observed in 1926, in Oslo and in a considerable number of localities in eastern Norway, and it is probable that the disease first appeared in that country in 1920. The disease has not yet been observed in Sweden or Denmark. It has not been found in the United States.

Very extensive damage has been caused by the disease in Holland, where Elms are found frequently throughout the country. In some towns, where these trees had been planted along the roadsides, the destruction has been very great, and hardly a healthy tree now remains. Several accounts of the disease have appeared in Germany, and recently it has attracted considerable attention in Norway, where a description of the disease and its occurrence in the country has recently been published in one of the daily papers at Oslo.

When trees are attacked, a mass of dry twigs and leaves appear in the crown, while the surrounding parts are yet green. The disease then gradually spreads over the whole tree and kills it. In young trees, from fifteen to twenty years old, the process usually takes a few weeks, but in some cases trees may lose their healthy appearance in three days, and the leaves may fall in the course of a week, leaving the tree quite bare. In trees from sixty to eighty years of age the disease spreads more slowly, and death may only take place after the lapse of several years. Such trees bud each year, but the leaf mass is thin and the leaves soon fall. Trees in which the twigs dry up early in the summer may produce fresh shoots, but these, in turn, wither. In winter, diseased trees can be recognized by the curved ends of the small twigs in the crown.

The disease has been found in the nursery where trees of four years and older are attacked. Young plants produced by layering have not been found infected.

In cross section diseased twigs and branches show a ring of brown spots just inside the bark. These are usually in the outermost annual ring, but in some cases may be found in the second and third ring from the outside. In severely diseased trees, every branch from the thickest to the smallest shows the discoloration of the wood and, in many cases, this may also be seen in the

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[1] Wilson, Malcolm. The Dutch elm disease. Gard. Chron. 81:133-134. Feb. 19, 1927.

roots. The bark of such trees is almost always riddled by the burrows of bark-beetles.

Almost all species of Ulmus appear to be attacked. The disease has been found on *Ulmus campestris latifolia, U. c. suberosa, U. monumentalis, U. hollandica, U. nitens* var. *ruepellii, U. americana* and *U. montana. U. vegeta* is said to be not attacked in Holland.

Up to the present the cause of the disease has not been definitely determined. No fungal hyphae can be found in the diseased branches, the discoloration being due to the deposition of a dark-brown granular substance in the vessels of the wood. Some investigators in Holland believe the disease to be due to the fungus *Graphium ulmi,* which has been obtained along with several other organisms from the diseased branches. Infection experiments with this fungus have, however, all given negative results.

It has also been suggested that the disease is due to physiological causes, especially the dry summer of 1921, and the cold winter of 1923-24, but as the disease is still spreading, this explanation seems inadequate. Recently, Brussoff, working in Germany, has stated that the disease is due to a bacterium, *Micrococcus ulmi,* which he has isolated from diseased trees. He states that he has carried out successful infection experiments with this organism on healthy Elm trees. Within the last year he has stated that the disease also attacks species of Acer, Tilia, Populus and Fagus, but he has not performed infection experiments on these trees. Although Brussoff's work does not appear to be altogether satisfactory, and requires confirmation, it is, up to the present, the best explanation that has been given.

As the disease is widespread in the four continental countries nearest to Britain, it is possible that, in spite of the prohibition of the importation of Elms, the disease may appear in this country. Any suspicious cases should, therefore, be at once submitted to mycological examination, so that, if the disease proves to be present, steps may be immediately taken to prevent its spread. Owing to its nature the disease is not one that can be easily identified and its presence can only be determined with certainty by a complete and thorough examination of the suspected trees.

In view of its very serious nature the Dutch authorities are now undertaking an extensive investigation of the disease in all of its aspects.

THE OCCURRENCE OF THE DUTCH ELM DISEASE IN ENGLAND

By MALCOLM WILSON and M. J. F. WILSON, Mycology Department, University of Edinburgh

An article dealing with the Dutch Elm Disease was published in *The Gardeners' Chronicle* in the early part of 1927 (February 19), in which the characters and distribution of the disease were shortly outlined. The disease was first discovered in South Holland, in September, 1919, and since that date has spread over the greater part of western Europe. Trees suffering from the disease may be readily recognized by the yellow discoloration of the leaves in the crown of the tree or at the tips of the side branches. This condition usually spreads rapidly over the tree and is followed by leaf-fall and, subsequently, by the death of the tree. Defoliation may be complete within a week, but sometimes extends over a much longer period. External symptoms of the disease are usually observed about the beginning of June. Sometimes a tree which has lost all its leaves early in the year may produce a second crop of leaves late in the season, but these are smaller and soon fall. Dying trees often produce large numbers of leafy shoots at the bases of the branches and on the main trunk. Infected branches, when cut across, show one or more rings of small brown spots in the most recently formed wood. If the bark is peeled from such twigs, longitudinal brown streaks are observed, which correspond to the brown spots in cross section. These internal symptoms can sometimes, but not invariably, be found in the roots.

As a consequence of the serious nature of the disease and its wide distribution on the Continent, orders have recently been made by the Ministry of Agriculture, and the Board of Agriculture for Scotland, prohibiting the importation of living Elm plants into Great Britain, in the hope of preventing the introduction of the disease. This hope, however, has not been realized, for an outbreak in the neighborhood of London was discovered last July. It is probable that the infection was already present in 1926, for in that year a number of Elms were reported to have died in the vicinity; these showed a discoloration of the leaves early in June, which was rapidly succeeded by leaf-fall and death of the tree. The trees were cut out, but, unfortunately, no examination was made, as at that time there was no suspicion of the presence of Dutch Elm Disease in Britain. In the early summer of 1927, another large tree died, showing similar symptoms, and, when felled shortly afterwards, showed the characteristic internal appearance of the disease.

During the investigation in July, it was observed that a neighboring tree showed discolored leaves in the crown; when examined in October, it was found that the discoloration had become intensified and had extended to almost all the branches on one side of the tree. All the infected branches, when cut across, showed the characteristic ring of brown spots in the wood, but this was absent in the roots.

Considering the severity and rapid spread of this disease, it is indeed remarkable that the cause of it should remain uncertain, this, too, in spite of the researches of several well-known pathologists both in Holland and Germany. There are at present three distinct opinions on the subject:—(1) That

the disease is caused by a fungus, *Graphium ulmi* Schwarz; (2) That the casual organism is a bacterium, *Micrococcus ulmi* Brussoff; (3) That the epidemic is not due to any parasite, but is a result of unfavorable weather conditions, such as drought or severe cold. The majority of the investigators, however, now consider that it is caused by the fungus *Graphium ulmi*. This fungus is characterized by the production of small coremia, which consist of a black stalk and enlarged head; the latter bears vast numbers of minute spores, which are embedded in a translucent drop of mucilage.

Specimens from the infected tree near London were carefully examined, and cultures were made from the discolored wood. *Graphium ulmi* developed in eighty per cent of the latter, but, in addition, bacteria appeared in a large proportion of the liquid cultures; most of these organisms were cocci, but rod-shaped bacteria were often present in small numbers. These results show a marked similarity with those obtained from authentic continental material. Further, the history of the death of these Elms near London, and the symptoms exhibited by the large trees examined there, are, in every detail, characteristic of the Dutch Elm Disease. These facts alone should be sufficient proof of the identity of the disease, but the evidence is strengthened by a recent publication by Dr. H. W. Wollenweber, a well-known mycologist in Berlin, whose infection experiments appear to prove conclusively that *Graphium ulmi* is the cause of the disease. The fact that this fungus was isolated in eighty per cent of the cultures made from British material is, therefore, particularly significant.

Some account of the damage caused by this disease on the Continent was given in the previous article, and later reports have emphasized the extremely serious nature of the epidemic. It is present throughout the whole of Holland and shows no sign of becoming less virulent; the situation in Belgium and in western Germany is almost similar. The Dutch authorities are attempting to remove the dead and most heavily infected trees, but they are unable to keep pace with the spread of the disease. In Holland, Elms have been largely used for planting on the dykes and in the towns, but, as a result of the disease, the planting of these trees is being discontinued.

No species of Ulmus planted in Holland appears to be immune to the disease, but the resistance to attack varies. Although it has been stated that other trees are liable to infection, there is no definite proof that the disease attacks any species outside the genus Ulmus.

Various methods of control have been attempted on the Continent. Autumn pruning of diseased branches has not been successful, and spraying has also proved ineffective. If the infection is at an early stage, and only isolated trees are attacked, removal and destruction by burning of the whole tree, including the roots, may check the spread of the disease.

Up to the present time there is only the one record in this country, and the infection, as yet, is not very severe, but, judging by the rapidity with which the disease has spread on the Continent, it is highly improbable that it will remain confined to this one area. If a calamity similar to that which has occurred in Holland is to be avoided, measures should be taken at once to control the disease, and for this purpose it is necessary to know whether there are any other outbreaks in the country. All suspected cases should, therefore, be reported without delay.